©2018 New Delhi Publishers. All rights reserved

MICROBIOLOGY

# Effect of Industrial Fly Ash on the Growth of Some Crop Field Soil Fungi Adapted with Ash Content

Shikha Shrivastava<sup>1</sup>, Pramod Kumar Mahish<sup>\*2</sup> and Anjali Ghritlahare<sup>3</sup>

<sup>1</sup>Department of Zoology, Indira Gandhi Govt. Arts and Commerce College Vaishali Nagar, Bhilai (Chhattisgarh) India <sup>2</sup>Department of Biotechnology, Govt. Digvijay (Autonomous) P.G. College Rajnandgaon (Chhattisgarh) India <sup>3</sup>Krishi Vigyan Kendra (KVK) Kanker, Affiliated to Indira Gandhi Agriculture University Raipur (Chhattisgarh) India

\*Corresponding author: drpramodkumarmahish@gmail.com (ORCID ID: 0000- 0002-5343-2347)

Paper No. 669

Received: 26-08-2017

Accepted: 16-01-2018

#### ABSTRACT

The study area of Chhattisgarh, known as rice bowl of India is rich in mineral resource. This has made cultivation of rice and coal-based iron and power industries run simultaneously. The industrial release fly ash comes to the cultivated soil. The microorganisms especially decomposers are very important with regard to the fertility of the soil. The present study therefore, focuses only to understand the effect of fly as on growth of some soil fungi. These fungi were isolated from crop field affected with the fly ash, while pure fly ash was added as media supplement obtained from power industry located in Korba. Simpson diversity index with 0.090 indicates a low diversity of fungi in the study area. *P. chrysogenum* has maximum growth (142.13%) in the low concentration (10 mg L<sup>-1</sup>) of fly ash followed by *C. lunata* and *A. niger*, while increased concentration showed mixed findings that support and also suppress the growth of some fungi. The ANOVA comparison (P = 0.035) therefore indicates the different effect of fly ash on fungi. From the findings it is concluded that the affected crop field soil has a low diversity of fungi but the still surviving may adopt it and be showing enhancement in growth while adding fly ash as nutrient.

#### Highlights

- A low biodiversity of fungi (Simpson index 0.090) was recorded in fly ash contaminated crop field.
- The isolated fungi tolerate high concentration of fly ash.
- The different ash concentration in the medium has different effect on fungal growth (P= 0.035).

Keywords: Fly ash, Soil property, Soil fungi, fly ash industry, biodiversity of fungi

Coal may be a standout amongst the major fossil fuels used for the generation of electricity in India. The coal-based thermal power plants use mostly bituminous coal with low calorific value. Furthermore high ash content leads to the production of the extensive volume of fly ash every year (Alam and Akhtar 2011). In the present situation, the whole nation is dependent on coal for power generation, this figure might increase further in the years to come. The respectable investigation has been focused on management and alternate use of fly ash in the manufacture of cement, bricks, landfilling, fertilizer fill and so on in constrained spot. These coal-based power plants generate enormous amount of fly ash (Meawad *et al.* 2010). The physical and chemical properties of coal ash are impacted towards coal source, moisture, particle size, furthermore for the kind of coal-burning process (Bhangare *et al.* 2011).

The utilization of fly ash for soil improvement has been advocated for three decades because of its ideal physico-chemical properties including appreciable amounts of K, Ca, Mg, S and P (Pandey *et al.* 2010a; Ram and Masto 2010). Fly ash contains generous amount of trace metals (Cu, Zn, Mn, and Mo) and toxic components such as vanadium (V), selenium (Se), arsenic (As), boron (B), aluminium (Al), Cd, lead (Pb), mercury (Hg) and Cr (Al-Ghouti



*et al.* 2011; Gupta *et al.* 2002). With this perspective, utilization of fly ash to improve agriculture productivity would not only be a solution to the problem but will also decline the utilization of inorganic fertilizers. This has great limitations because of the high toxic concentration of certain elements and heavy metals clinched alongside fly ash (Pandey and Singh 2010).

Most of the studies on fly ash are centered around its effect on plant development and productivity, heavy metal accumulation in plant and management practices to minimize the unfavorable effects of fly ash. However, the effect of fly ash on soil fertility, soil microbial/biochemical activity and soil nitrogen cycling is exceptionally restricted (Pandey and Singh, 2010). Microorganisms vary in their reaction to the rate of fly ash application. In a study, population of fungi and actinomycetes diminished with the use of fly ash (Nayak et al. 2014) since changes in microbial populations and metabolic activities can be identified even before the change in soil physico-chemical parameters. So, they might be acceptable and on time-pointer about soil degradation or improvement (Pankhurst et al. 1995). Soil microbes, especially mycorrhizal and saprophytic fungi, beneficial bacteria such as Rhizobium and plant growth promoting rhizobacteria (PGPR) acts as a major part in soil activities (Miransari and Mackenzie 2011). Arbuscular mycorrhizal fungi (AMF) promotes plant growth, encourages supplement take-up and enhances plant resistance to unfriendly conditions (Kaya et al. 2003; Labidi et al. 2012). On the other hand, saprophytic fungi (decomposers) have a fundamental function in soil and plant litter nutrient cycling in terrestrial ecosystems (Hättenschwiler et al. 2005). So, the present study aims to find out the effect of fly ash on the growth of some fungi isolated from ash contaminated soil.

# MATERIALS AND METHODS

The fly ash was collected from bricks manufacturing industry situated near the agriculture land site. The fly ash was collected from the surface of the site and soil sample was collected from a depth of 5-10cm. The collection site is situated 4 km from the laboratory of research centre (Department of Biotechnology, Govt. Digvijay College Rajnandgaon). The fly ash and soil sample were brought to laboratory into the sterile plastic bottles The microorganisms were isolated from soil using serial dilution method in which 1g soil sample was diluted to 10ml distilled water and diluted serially to  $10^{-1}$ ,  $10^{-2}$ ,  $10^{-3}$ ,  $10^{-4}$  and  $10^{-5}$ . The microbes were isolated from  $10^{-5}$  dilution on the agar plate. The pure cultures were maintained in agar slant for further use. Isolated fungi were mounted with Lactophenol cotton blue and observed under microscope (Binocular compound microscope made by labomade). The fungi were identified using available literature (Nagamani *et al.* 2006; Ellis 1971; Barnett and Hunter 1998). The isolated fungal species were subjected to find its diversity. Percent frequency was recorded and diversity index was recorded using Simpson and Shannon methods.

The effect of fly ash on the growth of some fungi was investigated using the supply of different concentration of fly ash on media supplement of fungi in 150ml conical flasks. A control was used to compare the value of test fungi. The concentration of 10, 100 and 1,000 mg L<sup>-1</sup> of fly ash was dissolved in vol/vol concentration of 50 ml Potato dextrose broth culture medium. Then the media was sterilized for 15 minutes at 15lbs pressure. After sterilization, 72 hr old fungal broth inoculum was transferred to medium. 1ml inoculum was aseptically transferred to each replicate. All the setup was maintained in triplicates. Then the flasks were incubated at 30°C for 7 days. The methodology was adopted from Mahish et al. (2015) with little modification replacing fly ash in the place of heavy metals. After incubation, mycelium was filtered using muslin cloth and allowed to (dry) in the oven for 8 hr at 80°C. Tolerance of fly ash by fungi was calculated in Tolerance Index (Ti) by measuring the Dry Weight (DW) of fungi (Fomina et al. 2005). The results were presented in tables and figure. The triplicate data was analyzed for mean and standard error. One way ANOVA was performed for the significance of the effect of fly ash on the growth of different fungi at P< 0.05.

# **RESULTS AND DISCUSSION**

In the present work, fungi were isolated from crop field contaminated with fly ash. The growth of fungi was tested against the different concentration of ash. The outcome of the work such as biodiversity of fungi, the growth of fungi from the different concentration of fly ash was presented with the help of tables, figures and the statistical summary.

### **Biodiversity of fungi**

Aspergillus niger (%F 19.04) was found as the dominant fungi followed by Curvularia lunata (%F 14.28), Peniciillium chrysogenum (%F 14.28), Penicillium sp. (%F 14.28) and Rhizopus sp. (%F 14.28). In total 21 colonies belonging to 09 species were detected. Diversity index was calculated and it was found as 0.090 (Simpson diversity index) with dominance index of 0.909, while Shannon diversity index was recorded as 3.01 (Fig. 1). The Simpson diversity index ranges from 0 to 1. Closer to 1 indicates high diversity and it is never negative. In the present study, 0.090 indicates a low diversity of fungi in the study area. The Shannon diversity represents the proportion of species abundance in the population. It is maximum when the all species occur in similar participation in diversity and is the lowest when the sample contains one species. So the high value represents the more diverse community. In the present study, value of 3.01 indicates the participation of all the species in the study area.

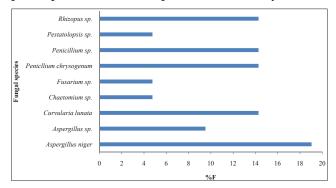


Fig. 1: Biodiversity of fungi from fly ash contaminated crop field soil

N=21, Simpson Diversity index = 0.090, Dominance index = 0.909; Shannon index = 3.01

#### Effect of fly ash on growth of some soil fungi

Based on the diversity, A. niger, Rhizopus sp., C.

*lunata, P. chrysogenum and Penicillium sp.* were screened for further study. These fungi were tested against 10, 100 and 1,000 mg L<sup>-1</sup> concentration of fly ash. Biomass of mycelium is presented in table 1. *P. chrysogenum* has maximum growth (142.13%) in the low concentration (10 mg L<sup>-1</sup>) of fly ash followed by *C. lunata* and *A. niger*. At increased concentration (100 and 1,000 mg L<sup>-1</sup> of fly ash) *P. chrysogenum, Rhizopus sp.,* and *A. niger* showed tolerance against fly ash, while least growth was observed in *Penicillium sp.* (Fig. 2.).

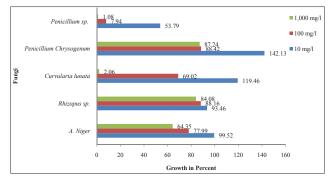


Fig. 2: Tolerance Index (Ti) of fungi in presence of different concentration of fly ash

The mean of the biomass obtained from all the fungi and from all the concentrations was subjected to one way ANOVA at significant level P 0.05. The P value was obtained 0.035, which is lower than the table value indication. There is significant difference between biomasses of fungi treated with fly ash in the medium supplement. The fly ash is generated from industries and spread as per the wind direction. The fly ash also gets deposited in the crop fields. The soil of the crop field may enrich by the deposition of fly ash or may pollute by huge deposition in continuation basis. It is greatly based on the chemistry of soil and growth of microorganism responsible for the solubility of nutrients (Pandey and Singh, 2010; Nayak et al. 2014). The present study dealt with the growth

**Table 1**: Growth of some soil fungi in different concentration of fly ash. Dry weight of fungal mycelium (Average<br/>of triplicate) Mean ± SE

Sl. No.	Concentration	A. niger	Rhizopus sp.	C. lunata	P. chrysogenum	Penicillium sp.
1	Control	$0.418 \pm 0.005$	$0.245 \pm 0.003$	$0.339 \pm 0.011$	$0.337 \pm 0.017$	$0.277 \pm 0.000$
2	10 mg L <sup>-1</sup>	$0.416\pm0.003$	$0.229 \pm 0.003$	$0.405\pm0.003$	$0.479 \pm 0.008$	$0.149\pm0.009$
3	100 mg L <sup>-1</sup>	$0.326 \pm 0.008$	$0.216 \pm 0.005$	$0.234\pm0.006$	$0.298 \pm 0.012$	$0.022 \pm 0.013$
4	1,000 mg L <sup>-1</sup>	$0.269\pm0.004$	$0.206 \pm 0.010$	$0.007\pm0.004$	$0.294\pm0.016$	$0.003 \pm 0.003$

ANOVA at P 0.05, F= 3.439, P= 0.035.



of soil fungi related to the fertility of crop field at different concentration of fly ash. The growth of *Penicllium chrysogenum* and *Curvularia lunta* was found more than the control at low concentration of ash (10 mg L<sup>-1</sup>) indicating the presence of enrichment of nutrients in the medium as previously recorded by Singh *et al.* (2012) and Parab *et al.* (2013).

## CONCLUSION

The economy of the country like India is principally in view of agriculture. The production of crop gets influenced by many reasons; ecological contamination is one of them. The present study reports a low biodiversity of soil microorganism is not a decent sign. While a few fungi may adopt on it and furthermore develop well in the low concentration of fly ash however, a high concentration was found diminished in the microbial development. So the use of small quantity of fly ash may enhance soil fertility, on the other hand, a higher concentration could be toxic to beneficial microorganisms.

## REFERENCES

- Alam, J. and M.N. Akhtar. 2011. Fly ash utilization in different sectors in Indian scenario. *Int. J. Emerg. Trends Eng. Dev.*, 1: 1–14.
- Al-Ghouti, M.A., Y.S. Al-Degs, A. Ghrair, H. Khoury and M. Ziedan. 2011. Extraction and separation of vanadium and nickel from fly ash produced in heavy fuel power plants. *Chem. Eng. J.*, **173**(1): 191–197.
- Amiri, F., Yaghmaei, S. and Mousavi, S.M. 2011. Bioleaching of tungsten-rich spent hydrocracking catalyst using *Penicillium simplicissimum. Bioresour. Technol.*, **102**(2): 1567–1573.
- Arivazhagan, K., Ravichandran, M., Dube, S., Mathur, V., Khandakar, R., Yagnanarayana, K., Kamal Pasha, M., Sinha, A., Sarangi, B., Tripathi, V., Gupta, S., Singh, R., Ali, M., Thakur, A. and Narayan, R. 2011. Effect of Coal Fly Ash on Agricultural Crops: Showcase Project on Use of Fly Ash in Agriculture in and Around Thermal Power Station Areas of National Thermal Power Corporation Ltd., India World coal Ash Conference, May 9–12, 2011, in Denver, CO, USA.
- Barnett, H.L. and Hunter, B. 1998. Illustrated Genera of Imperfect fungi. APS Press, St Paul, Minnesota, p. 218.
- Bhangare, R., Ajmal, P., Sahu, S., Pandit, G. and Puranik, V. 2011. Distribution of trace elements in coal and combustion residues from five thermal power plants in India. *International Journal of Coal Geology*, 86: 349–356.
- Ellis, M.B. 1971. Dematiaceous Hyphomycetes. CAB International, Oxon, UK.

- Fomina, M.A., Alexander, I.J., Colpaert, J.V. and Gadd, G.M. 2005. Solubilization of toxic metal minerals and metal tolerance of mycorrhizal fungi. *Soil Biol. Biochem.*, 37: 851-866.
- Garcia –Sanchez, M., Garcia-Romera, I., Cajthaml, T., Tlustos, P. and Szakova, J. 2015. Changes in soil microbial community functionality and structure in a metal-polluted site: The effect of digestate and fly ash applications. *Journal* of Environmental Management, **162**: 63-73
- Giller, K.E., Witter, E. and McGrath, S.P. 2009. Heavy metals and soil microbes. *Soil Biol. Biochem.*, **41**: 2031-2037.
- Gupta, D.K., Rai, U.N., Tripathi, R.D. and Inouhe, M. 2002. Impacts of fly ash on soil and plant responses. *J. Plant Res.*, **115**: 401–409.
- Hättenschwiler, S., Tiunov, A.V. and Scheu, S. 2005. Biodiversity and litter decomposition in terrestrial ecosystems. Annu. *Rev. Ecol. Evol. Syst.*, **36**: 191–218.
- Jadhav, U.U. and Hocheng, H. 2015. Analysis of Metal Bioleaching from Thermal Power Plant Fly Ash by *Aspergillus niger* 34770 Culture Supernatant and Reduction of Phytotoxicity During the Process. *Appl. Biochem. Biotechnol.*, **175**: 870–881.
- Jala, S. and Goyal, D. 2006. Fly ash as a soil ameliorant for improving crop production a review. *Bioresour. Technol.* 97: 1136-1147.
- Kaya, C., Higgs, D., Kirnak, H. and Tas, I. 2003. Mycorrhizal colonisation improves fruit yield and water use efficiency in watermelon (*Citrullus lanatus* Thunb.) grown under well-watered and water-stressed conditions. *Plant Soil*, 253: 287–292.
- Labidi, S., Ben Jeddi, F., Tisserant, B., Debiane, D., Rezgui, S., Grandmougin-Ferjani, A. and Lounès-Hadj Sahraoui, A. 2012. Role of arbuscular mycorrhizal symbiosis in root mineral uptake under CaCO<sub>3</sub> stress. *Mycorrhiza*, 22: 337–345.
- Mahish, P.K., Tiwari, K.L. and Jadhav, S.K. 2015. Biodiversity of Fungi from Lead Contaminated Industrial Waste Water and Tolerance of Lead Metal Ion by Dominant Fungi. *Research Journal of Environmental Sciences*, 9(4): 159-168.
- Meawad, A., Bojinova, D. and Pelovski, Y. 2010. An overview of metals recovery from thermal power plant solid wastes. *Waste Management*, **30**: 2548–2559.
- Miransari, M. and Mackenzie, A.F. 2011. Development of a soil N-test for fertilizer requirements for corn (*Zea mays* L:) production in Quebec. *Comm. Soil Sci. Plant Anal.*, **42**: 50–65.
- Nagamani A., Kunwar, I.K. and Manoharachary, C. 2006. Handbook of soil fungi. IK International, New Delhi.
- Navarro, R., Guzman, J., Saucedo, I., Revilla, J. and Guibal, E. 2007. Vanadium recovery from oil fly ash by leaching, precipitation and solvent extraction processes. *Waste Manage.*, **27**(**3**): 425–438.
- Pankhurst, C.E., Hawke, B.G., McDonald, H.J., Kirkby, C.A., Buckerfield, J.C., Michelsen, P. and Doube, B.M. 1995. Evaluation of soil biological properties as potential bioindicators of soil health. *Anim. Prod. Sci.*, 35: 1015–1028.



- Pandey, V. and Singh, N. 2010. Application of fly ash on the growth performance and translocation of toxic heavy metals within *Cajanus cajan* L.: Implication for safe utilization of fly ash for agricultural production. *Agriculture Ecosystems & Environment*, **136**: 16–27.
- Pandey, V.C. and Singh, N. 2010a. Impact of fly ash incorporation in soil systems. *Agric. Ecosyst. Environ.* 136: 16–27.
- Pandey, V.C., Singh, J.S., Kumar, A. and Tewary, D.D. 2010b. Accumulation of heavy metals by chickpea grown in fly ash treated soil: effect on antioxidants. *Clean Soil Air Water*, **38**: 1116–1123.
- Parab, N., Mishra, S. and Bhonde, S.R. 2013a. Azotobacter chroococccum: a potential organism in the management of crop yield and quality under fly ash amended soil. *Int. Soc. Environ. Bot. Newslett.*, **19**(1): 6–8.
- Parab, N., Mishra, S. and Bhonde, S.R. 2013b. Mycorrhizal dependency, yield and biochemical parameters of onion in response to fly ash amendment with different biofertilizers. *Int. J. Environ. Sci.* **2**(**1**): 34–41.
- Parab, N., Sinha, S. and Mishra, S. 2015. Coal fly ash amendment in acidic field: Effect on soil microbial activity and onion yield. *Applied Soil Ecology*, **96**: 211–216.
- Qu, Y., Lian, B., Mo, B. and Liu, C. 2013. Bioleaching of heavy metals from red mud using *Aspergillus niger*. *Hydrometallurgy*, **136**: 71–77.

- Ram, L.C. and Masto, R.E. 2010. Review: an appraisal of the potential use of fly ash for reclaiming coal mine spoil. *J. Environ. Manag.*, **91**: 603–617.
- Rizvi, R. and Khan, A. 2009. Response of eggplant (*Solanum melongena* L.) to fly ash and brick kiln dust amended soil. *Biol. Med.*, **1**(2): 20–24.
- Singh, A., Sarkar, A. and Agrawal, S.B. 2012. Assessing the potential impact of fly ash amendments on Indian paddy field with special emphasis on growth, yield and grain quality of three rice cultivars. *Environ. Monit. Assess.*, 184(8): 4799–4814.
- Stefanowicz, A.M., Niklinska, M., Kapusta, P. and Szarek-Łukaszewska, G. 2010. Pine forest and grassland differently influence the response of soil microbial communities to metal contamination. *Sci. Total Environ.*, 408: 6134-6141.
- Tordoff, G.M., Baker, A.J.M. and Willis, A.J. 2000. Current approaches to the revegetation and reclamation of metalliferous mine wastes. *Chemosphere*, **41**: 219-228.
- Xu, T.J. and Ting, Y.P. 2009. Fungal bioleaching of incineration fly ash: metal extraction and modeling growth kinetics. *Enzyme Microb. Technol.*, 44 (5): 323–328.